SUMMARY STATEMENT

PROGRAM CONTACT: [redacted]

(Privileged Communication)

Release Date: **12/07/2016**

Revised Date:

Application Number: **2 R01 DC012557-06**

Principal Investigator

FROEMKE, ROBERT CROOKS

Applicant Organization: [redacted]

Review Group: **ZRG1 AUD-Z (90)**
Center for Scientific Review Special Emphasis Panel
Auditory System Special Review

Meeting Date: **11/07/2016**
Council: **JAN 2017**
Requested Start: **04/01/2017**

RFA/PA: **PA16-160**
PCC: **HR61**

Dual IC(s): **MH, LM**

Project Title: **Synaptic basis of perceptual learning in primary auditory cortex**

SRG Action: **Impact Score:11 Percentile:1 &**
Next Steps: **Visit http://grants.nih.gov/grants/next_steps.htm**
Human Subjects: **10-No human subjects involved**
Animal Subjects: **30-Vertebrate animals involved - no SRG concerns noted**

Project Year		
6	[redacted]	[redacted]
7		
8		
9		
10		

ADMINISTRATIVE BUDGET NOTE: The budget shown is the requested budget and has not been adjusted to reflect any recommendations made by reviewers. If an award is planned, the costs will be calculated by Institute grants management staff based on the recommendations outlined below in the COMMITTEE BUDGET RECOMMENDATIONS section.

2R01DC012557-06 FROEMKE, ROBERT

RESUME AND SUMMARY OF DISCUSSION: The work proposed in this R01 renewal application is designed to examine the role of the cholinergic and noradrenergic neuromodulatory systems during the regulation of synaptic plasticity in the auditory cortex during auditory perceptual learning. Sophisticated methods will be used, including optogenetics, two-photon imaging and electrophysiological recordings in behaving mice. The panel considered this work of very high significance given the importance of the modulatory systems in question, the limited understanding and study of them in this context, and the prospect for this work to make important discoveries relevant to both the basic and translational science of the auditory system. Other noted strengths included the outstanding investigator, the technically impressive methods, the rigorously designed research strategy, with a strong scientific premise and ample preliminary data, “astounding productivity” in the prior funding period, and the fact that this work is a very logical extension of the studies in the prior funding period. No significant weaknesses were discussed, and panel widely agreed that the proposed work will lead to very high impact advances relevant to the regulation of synaptic plasticity during auditory learning.

DESCRIPTION (provided by applicant): The brain has the remarkable capability to change in response to experience. This plasticity is essential for learning and memory, and is an important feature of the auditory cortex, especially for learning the significance of sensory signals such as speech, for the use of devices such as cochlear implants, and for recovery after short- term deafness. These changes are thought to occur primarily at synapses, basic units of information processing and plasticity. Long-term synaptic plasticity requires sensory experience and activation of neuromodulatory systems which convey behavioral context to local cortical circuits. However, little is known about the interactions between synaptic inputs and release of neuromodulators in vivo, making it challenging to relate perceptual learning to plasticity in the auditory cortex or other brain areas. Recently we developed an approach to measuring dynamics of synaptic modifications for hours, coupled with imaging techniques enabling us to monitor the same cells over days during training, directly monitoring and manipulating activity in behaving mice. These approaches allow for a close examination of links between modulation, cortical plasticity and auditory perceptual learning. Specifically, we will study how auditory perceptual training activates the cholinergic vs noradrenergic modulatory systems. These two modulators are principally involved in selective attention towards behaviorally-important stimuli, general arousal, and learning. However, there may be important functional differences in these systems in terms of when they are active during different phases of training or consequences of cholinergic and noradrenergic modulation on auditory neurons for contextual information processing. This proposal describes a series of imaging, recording, optogenetic, and behavioral experiments that will compare and contrast the effects of locus coeruleus activation and norepinephrine release vs the effects of nucleus basalis activation and cholinergic modulation on the primary auditory cortex of behaving mice. Many studies have highlighted the importance of recording in awake animals during behavior, and we will first examine how ensembles of excitatory and inhibitory neurons are affected by learning over the entire duration of training, as animals go from naïve and poor performers, to having reliable performance on an auditory detection and recognition task we have used in the lab for years. Next, we determine when and how cholinergic and noradrenergic modulation affect behavioral and neural responses. Finally, we will make some of the first direct measurements of modulatory neuron responses, asking how these systems are activated by task-relevant variables such as sounds linked to reward. In summary, here we use in vivo recording and imaging methods to ask how behavioral training engages and modifies noradrenergic and cholinergic systems, to collectively affect auditory cortical processing and persistently improve auditory perceptual abilities in behaving mice.

PUBLIC HEALTH RELEVANCE: Neuroplasticity- the ability of the brain to change in response to experience- is an essential feature of the auditory cortex, especially for speech and language learning as well as the successful use of devices such as cochlear implants. However, it is unclear how motivational state and behavioral training drive plasticity within the central auditory system. The experiments to be performed in this proposal provide essential data on basic mechanisms of

neuromodulation and plasticity in the auditory cortex, required for improvement of prosthetic design and therapeutic strategies for treatment of deafness and language disorders.

Critique 1

Significance: 1
Investigator(s): 1
Innovation: 1
Approach: 2
Environment: 1

Overall Impact: The cholinergic and noradrenergic systems are clearly important modulators of neural activity, acting directly at the synapse and being heavily involved in learning. Auditory learning is important, particularly in early human life, and yet the effect of these neuromodulators in auditory cortex receives precious little attention. The proposal is a continuation in Dr. Froemke's study of the effect of neuromodulators on learning and plasticity in the auditory pathway. The results obtained in this study, as well as during his previous grant cycle, are bound to have a major influence on our view of the role of motivation and environment in language acquisition, language disorders as well as the retraining necessary following cochlear implantation.

1. Significance:

Strengths

- This proposal builds on results and on-going research from the investigator's previous RO1 grant on the same subject.
- *Impressive* amount of research was conducted, and results published, over the last few years, with more results coming.
- As mentioned in the previous application, and still very much true, the studies of the cholinergic and noradrenergic systems complement existing studies of modulation of activity in auditory cortex (including top-down, circuit modulation), adding important new information about the role and importance of these neuromodulatory transmitters.

Weaknesses

- None really. The proposal was quite dense and a bit hard to read at times. Fewer details sometimes make points easier to grasp.

2. Investigator(s):

Strengths

- Impressive publication record and productivity in general.

Weaknesses

- None

3. Innovation:

Strengths

- Many modern techniques are used. Their use is always justified. This is a difficult subject with comparatively little background to go on, and Dr. Froemke does an outstanding job at disentangling the relative contributions of the different neuromodulators under study.

Weaknesses

- None

4. Approach:

Strengths

- Systematic, thorough and well informed of previous research, including in related fields.
- Ongoing publication record speaks best as an answer to this question!

Weaknesses

- Proposal is quite dense and a bit hard to follow at times
- Minor: All drugs are applied bilaterally but is there laterality of some of the effects? How do both auditory cortices keep synchrony in effect size, for example.

5. Environment:

Strengths

- NYU School of Medicine has all the resources necessary to accomplish the work proposed.

Weaknesses

- None

Protections for Human Subjects:

Not Applicable (No Human Subjects)

Vertebrate Animals:

YES, all four points addressed

- This is well justified, with much more details than are usually found.
- Both sexes will be used, but sex as a variable should be addressed more explicitly.

Biohazards:

Acceptable

Renewal:

- The very impressive body of published work that arose from the first 5 years of this grant speaks volumes about the proposer's ability to carry work to completion.

Resource Sharing Plans:

Acceptable

Authentication of Key Biological and/or Chemical Resources:

Acceptable

Budget and Period of Support:

Recommend as Requested

Critique 2

Significance: 2
Investigator(s): 1
Innovation: 2
Approach: 1
Environment: 1

Overall Impact: This is a proposal for continuation of a successful research program on neuromodulation in auditory cortex plasticity. The reviewers found the proposed research to be a logical continuation of previous work, well justified by current knowledge and precisely calibrated to advance our understanding of one of the most interesting problems in neuroscience, the role of neural plasticity in learning and the relationship between behavioral learning and brain mechanisms. The proposal is well developed and exciting.

1. Significance:

Strengths

- The proposed work is directed at questions of considerable current interest, for both basic and translational science.
- The proposal is a natural evolution of the ideas and results derived from the PI's previous work, carrying the analysis from phenomena to mechanisms.
- The proposal builds on a strong foundation of research on the role of neuromodulators in learning, demonstrated by behavioral and neurophysiological research. Thus the ideas are sharply focused and there are clear and testable hypotheses.

Weaknesses

- None noted

2. Investigator(s):

Strengths

- Froemke is a very successful early-to-mid career neuroscientist. He has extensive experience with all the research tools needed for this work.
- The PI's laboratory has been very productive over the first 5 years of this grant support.

Weaknesses

- None

3. Innovation:

Strengths

- The techniques to be used here are state of the art in cortical neurophysiology. The PI has marshaled the most powerful and appropriate combination of methods for each of the aims of the proposal.
- The proposal to directly record in NB and LC is an important addition to the analysis of cortical plasticity.

Weaknesses

- None noted

4. Approach:

Strengths

- The planned experiments are a systematic and logical sequence proceeding from the properties of cortical neurons, to the dependence of those properties on the ACh and NE systems, to the response characteristics of the neuromodulatory source neurons.
- The ability to study neurons with both intracellular (whole-cell) and population (2-photon) recording will yield a comprehensive view of the neural activity.
- The plan to separate excitatory from inhibitory inputs in whole-cell recording is a powerful approach to analysis of synaptic plasticity.
- Preliminary data are provided to show that the proposed experiments can be successfully done in the PI's laboratory.
- Multiple technical approaches are planned for most of the aims. Thus, if something doesn't work, there's usually another approach in reserve.
- Appropriate plans for statistical testing are given in the methods and the number of animals to be used is determined on the basis of the expected statistical power of the data.

Weaknesses

- None noted

5. Environment:

Strengths

- The PI's laboratory is well-equipped. Most of the equipment needed for the proposed work is contained in the lab or in the PI's department. Some needed equipment and upgrades will be added in year 3.
- The [redacted] Institute is an excellent environment for the proposed work with good colleagues for support and scientific discussions. The PI seems to be well-mentored.

Weaknesses

- None noted

Protections for Human Subjects:

Not Applicable (No Human Subjects)

Vertebrate Animals:

YES, all four points addressed

- The use of mice and the animal numbers is well justified by the proposed work. Both male and female animals will be used. There is adequate veterinary care available and the procedures to minimize discomfort are appropriate. The euthanasia plan is satisfactory.

Biohazards:

Not Applicable (No Biohazards)

Renewal:

- Progress during the previous grant period has been very good. The previous work lays an excellent foundation for the work proposed here.

Authentication of Key Biological and/or Chemical Resources:

Acceptable

Budget and Period of Support:

Recommend as Requested

Critique 3

Significance: 2
Investigator(s): 1
Innovation: 2
Approach: 1
Environment: 2

Overall Impact: This is a well-written renewal that proposes a logical extension of some very successful studies focused on plasticity in auditory cortex. The underlying premise is that acetylcholine and noradrenaline modulate auditory cortex during contextual learning. The new studies will use highly innovative optogenetic and imaging methods in awake animals to monitor cholinergic and noradrenergic modulation of task-related plasticity in primary auditory cortex. The results will move the field forward regarding the relative contributions of acetylcholine and norepinephrine in controlling the plasticity of excitatory and inhibitory cortical neurons during different phases of learning an auditory task.

1. Significance:

Strengths

- These experiments promise a unique opportunity to dissect the differential contributions of acetylcholine and noerepinephrine to inter-related contributions to plasticity and learning of an auditory task.

Weaknesses

- None

2. Investigator(s):

Strengths

- The PI has been a leader in developing many of the techniques to be used in the current studies.

Weaknesses

- None

3. Innovation:

Strengths

- Simultaneous examination of ACh and NE axon activity in primary auditory cortex.

Weaknesses

- None

4. Approach:

Strengths

- Rigor is addressed appropriately. Both sexes will be used. Strains are discussed appropriately. Group sizes are determined appropriately.
- The proposed work forms a logical extension of previous work in that it looks at NE and ACH simultaneously; 2) it looks at effects in un-anesthetized animals.
- Substantial preliminary data suggests shows a high likelihood for success of the experiments.
- The 3 Aims represent independent but highly related experiments that together produce a logical set of results that will move the field forward regardless of the particular findings.

Weaknesses

- Two-photon imaging is largely limited to the superficial layers of cortex, preventing the experimenters from collecting data from neurons in the deeper layers in certain experiments. The advantages of this approach probably outweigh the limitations, but the PI acknowledges the compromise.

5. Environment:

Strengths

- NYU provides an outstanding intellectual environment
- All necessary equipment is available, with local expertise in abundance for troubleshooting any problems that might arise

Weaknesses

- None

Protections for Human Subjects:

Not Applicable (No Human Subjects)

Vertebrate Animals:

YES, all four points addressed

Renewal:

- The PI has been highly productive during the last grant period.
- The proposed experiments represent a logical extension of the previous work.

Budget and Period of Support:

Recommend as Requested

THE FOLLOWING SECTIONS WERE PREPARED BY THE SCIENTIFIC REVIEW OFFICER TO SUMMARIZE THE OUTCOME OF DISCUSSIONS OF THE REVIEW COMMITTEE, OR REVIEWERS' WRITTEN CRITIQUES, ON THE FOLLOWING ISSUES:

VERTEBRATE ANIMAL (Resume): ACCEPTABLE

COMMITTEE BUDGET RECOMMENDATIONS: The budget was recommended as requested.

Footnotes for 2 R01 DC012557-06; PI Name: FROEMKE, ROBERT CROOKS

& Ad hoc or special section application percentiled against 2017/01 AUD study section.

NIH has modified its policy regarding the receipt of resubmissions (amended applications). See Guide Notice NOT-OD-14-074 at http://grants.nih.gov/grants/guide/notice-files/NOT-OD-14-074.html. The impact/priority score is calculated after discussion of an application by averaging the overall scores (1-9) given by all voting reviewers on the committee and multiplying by 10. The criterion scores are submitted prior to the meeting by the individual reviewers assigned to an application, and are not discussed specifically at the review meeting or calculated into the overall impact score. Some applications also receive a percentile ranking. For details on the review process, see http://grants.nih.gov/grants/peer_review_process.htm#scoring.